Salmonella bacteremia with confirmed mycotic aneurysm at surgery or autopsy.

Results (see table): Overall, the most common species were *Salmonella typhimurium* (26%), *Salmonella enteritidis* (24%) and *Salmonella choleraesuis* (20%).

Conclusions: An important risk factor for SA is atherosclerosis. Due to the high mortality of SA, patients with atherosclerosis and Salmonella enterocolitis should be treated with quinolones even without documented bacteremia. In case of fever, blood cultures should be performed after stopping antibiotics, in order to detect recurrent bacteremia as a sign of aortitis.

P576 Is an Infection by *Chlamydia pneumoniae* a Risk Factor for a Cardiovascular Endpoint?

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Objectives: This prospective study tries to find out a link between an acute or chronic infection by *Chlamydia pneumoniae* and a cardiovascular event.

Methods: Serological tests of 58 patients and of 58 controls (age and sex matched) were analyzed for recent or past infections of *Chlamydia pneumoniae*. The second serological test was done 4 to 6 weeks later. Titers of IgG, IgM and IgA were determined by Microtiter indirect immunofluorescence test. The patients had suffered an acute myocardial infarction or a coronarographic proven angor in the week preceding the first serological examination.

All patients and controls were asked about their cardiovascular risk factors and about an upper respiratory tract infection during 6 weeks before their hospitalization.

Results: Patients with a coronary event had statistically significantly more histories of upper tract infections than controls ($p = 0.03$). 19/58 patients and 24/58 controls had an serologically proven infection (IgG, IgA, IgM) by *Chlamydia pneumoniae* ($p = 0.93$). Only 2 patients and 1 control had an acute infection by *Chlamydia pneumoniae*. This study could not confirm that an infection by *Chlamydia pneumoniae* is a risk factor for a cardiovascular event. The prevalence of positive serologies of *Chlamydia pneumoniae* in patients and controls were the same, and were similar to the prevalence in Europe. Our results don't confirm the findings of other studies. We think that a serological analysis alone might not be sensible enough to detect all upper respiratory tract infections by *Chlamydia pneumoniae*. It is likely that a PCR-test could give more information. Furthermore, other infectious agents may play a role in the pathogenesis of an acute cardiovascular event.

P577 Seroprevalence of Chlamydia pneumoniae in Coronary Heart Diseases

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Objective: The relationship of *Chlamydia pneumoniae (C. pneumoniae)* to coronary heart diseases and the generation of atherom plaques in coronary arteries has been suggested by seroepidemiologic studies and by observation of microorganisms in atheriosclerotic lesions by direct methods.

Methods: In this study, we investigated 58 serum samples of patients with coronary heart disease who undergone coronary angiography and a control group of 20 healthy subjects who were selected from asymptomatic patients. The investigation was made using the indirect immunofluorescent antibody assay (IIFA) in order to detect a possible relationship between coronary heart diseases and *C. pneumoniae* infections. The aim of this study was to determine seroprevalence of *C. pneumoniae* in coronary heart diseases. We accepted 1/100 and higher titers as seropositive. We determined whether or not patients with coronary heart disease had angina pectoris, myocardial enfarctus, hypertension and diabetes mellitus as possible risk factors.

Results: We found that 41 (75.9) patients with coronary heart disease symptoms were seropositive for C. pneumoniae and 10 (50%) in the control group. In 25 (54.4%) of the patients with coronary heart disease, *C. pneumoniae* infection was the only risk factor.

Conclusion: Although our findings suggest a positive relationship between *C. pneumoniae* and atheriosclerosis as other studies have shown, and since specific antimicrobial therapy for atheriosclerosis for these patients is not yet being used, it is important that physicians become aware of this relationship.

P578 Coronary Arteries Harbour Viable *Chlamydia Pneumoniae*

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Objectives: Indirect seroepidemiologic evidence suggests former infection with the intracellular bacterial pathogen *Chlamydia pneumoniae* to be a risk factor for coronary heart disease and acute myocardial infarction. This investigation was made to ensure recovery of viable *C. pneumoniae* from atheromatous plaques of stenotic human coronary arteries.

Methods: Coronary endarterectomy samples were examined for presence of genomic *C. pneumonia* DNA in a nested PCR ($n = 120$) and for the presence of viable chlamydiae by cell culture ($n = 60$). Patient sera were examined by a microimmunofluorescence assay.

Results: Viable, continuously replicative *C. pneumoniae* were recovered from 8% of atherosclerotic plaques. 24% of the coronary samples were positive for chlamydial DNA. Infection appeared limited to progredient atherosclerotic lesions. There was no apparent histologic distinction between infected and non-infected tissue. Serology was of no use in identifying the patients with endovascular infection.

Conclusions: Results demonstrate a substantial part of atherosclerotic coronary arteries to be infected with viable *C. pneumoniae*. A causal contribution of the endovascular infection to atherogenesis and coronary heart disease remains to be established.

Brucella, diphtheria, leptospira

P579 Microtechnique of 2-Mercaptoethanol Test in Serologic Diagnosis of Human Brucellosis

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Objectives: A comparative assessment of Miotechnique of 2-Mercaptoethanol test (MAT 2-ME) in comparison with 2-Mercaptoethanol test (2-ME) in serologic diagnosis of human brucellosis.

Methods: 2-ME was performed in tubes and MAT 2-ME in microplates. The sera were incubated at 37° C for 24 h. Titers ≥ 160 were considered as positive. These tests confirm only IgG as agglutinable antibodies in sera.

Results: A total of 223 sera were examined with both methods. Equal titers were in 212 (95%). The differences of two or more dilutions in 11 (5%) sera were statistically insignificant ($p > 0.05$).

MAT 2-ME has some advantages: need 20 fold lesser *Brucella* antigen, the irritation of mucoses is significantly lesser, the simply and fast use, the additional equipment is not expensive.

Conclusions: MAT 2-ME is very accurate method with some advantages in comparison with 2-ME. Since the results of this test, in combination with the results of The Serum Agglutination Test-Wright, are an important indicator of the activity and the stage of the disease and reponse to the antibiotic treatment, this test should be used as one of the routine serologic tests.

P580 Diagnosis of Human Brucellosis by A Single Tube Nested Polymerase Chain Reaction Assay

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Objectives: To develop a single tube nested polymerase chain reaction (PCR) assay for detection of *Brucellae* in human blood.

Methods: A single tube nested PCR was developed with outer and inner primers derived from IS711, a gene common to all *Brucellae*. The PCR products were detected by colorimetry. The test was applied in field conditions with blood specimens from 28 clinically diagnosed brucellosis patients, 28 patients with fevers due to other causes, and 28 healthy controls.

Results: The single tube nested PCR gave positive reactions with 14 strains of five *Brucellae* species, and detected as few as 30 organisms. There were no false positive PCR reactions with a range of bacteria known to evoke serological cross-reactions with *Brucellae*. Blood samples from 28 subjects with clinically suspected brucellosis gave a positive PCR. Samples from 28 patients with fever of other causes, and samples from 28 healthy controls gave negative PCR results.

Conclusion: The established single tube nested PCR could be a valuable method in the specific diagnosis of human brucellosis.

Acknowledgment: This study was supported by KU Grant M1085.

P581 Seroprevalence of Brucelosis

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Portugal, as a mediterranean country with tradition of husbandry, presents a high Brucellosis incidence.

Objectives: To know the Brucellosis seroprevalence in a endemic area and to compare laboratorial tests (Rose Bengal-RB, ELISA, Imunofluorescent Assay-IFA).

Methods: Epidemiological and serological case-control study in 346 rural inhabitant, randomly select from two similar communities. Data were gathered using a questionnaire and by blood collecting. All the serum were tested by RB and ELISA. The positive serum were tested by IFA. It was done another blood collecting after 6 months only on the persons who have had positive tests or Brucelosis before.

Results: In the control population, two IgA positive serum were found with ELISA but one of them was from a previously detected Brucellosis case. In the case population, a lower rate of positive serum (1.48%) was found using RB. With ELISA it was found 0.74% IgM positive, 7.35% IgG positive and 9.56% IgA positive. Using IFA, 5.88% IgG positive serum was detected.

Conclusions: In the endemic community, we found a Brucellosis seroprevalence of 12.50% which is similar to another studies. With regard to laboratory tests we found that there is no correlation between RB and ELISA. Between ELISA and IFA the correlation is positive ($r^2 = 0.32$).

We found the populations screenings not effective because the cost/benefit relation isn't positive. We think that developing educational/information to country people and increase the epidemiological surveillance in endemic communities with laboratorial screenings tests like ELISA or IFA will be more effective measures to take in terms of public health.

P582 Childhood Brucellosis in Southern Spain

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Objectives: To describe the epidemiology, clinical features, laboratory findings and outcome of pediatric brucellosis in Southern Spain.

Methods: Retrospective chart review of patients younger than 14 years of age diagnosed of having brucellosis between 1985 and 1995.

Results: Sixty three patients, ages ranging from 18 months to 13 years, had brucellosis. Fever (100%), sweating (50.8%) and arthralgias (49.2%) were the commonest presenting symptoms. Liver enzymes were elevated in 62 percent of the patients. *Brucella Mellitensis* was identified in 80.3 percent of the cases. Eleven patients (17.5%) had focal complications. Peripheral arthritis was present in seven patients. Another two additional patients had axial skeleton involvement. There was one case of neurobrucellosis and a child presented evidence of DIC. The duration of symptoms before diagnosis was similar in patients with uncomplicated courses to those with focal complications (13.79 ± 9.16 vs 14.92 ± 6.14). Thirty eight patients received doxycycline for 3 (30) or more weeks (8) plus streptomycin for 2 weeks. Eighteen children were treated with rifampin either, for 3 (7) or 4 (11) weeks along with streptomycin. Remaining patients were given different combined therapy schedules. The overall relapse rate was 11.1%. There were no relapses among the patients treated with rifampin plus streptomycin. Four patients (13.3%, CI 3.16–23.50%) who received 3 weeks of doxycycline plus streptomycin relapsed.

Conclusions: Brucellosis remains a significant health problem among children in Southern Spain. Three weeks of doxycycline plus streptomycin may not be fully effective in preventing relapses.

P583 Rucellosis-Clinical and Epidemiological Characteristics in Our Region

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This work is a review of the registered patients suffuring from Brucellosis in Bitola and its surrounding villages in the period of 1980 to 1990. 398 patients were hospitalized with an average duration of the hospitalization of 21 days. 93.69% were completely cured, 6.03% were relapses. According to their profession 53.26% were cattle breeders; 28.6% housewives; 13.32% students; 1.26% veterinarians; 4.27% rest. The dominant way of infection was the direct contact with the infected cattle. According to age: 40.2% belonged to the group of 40 to 60 years of age; 32.66% to the group od 20 to 40 years; 20.1% to the group of 0 to 20 years, and 7.8% belonged to the group over 60 years. According to the symptoms dominance, patients were grouped: patients with symptoms on the locomotor system with 28.64%; patients with damage of visceral organs with 39.6%; with liver damage 15.07%; with orchiepydidymitis 4.52%; with limphadenopaties 19.85% and with general infectious syndrom

23.60%. According to our clinical material the biggest number (148) were registered in 1981. In the following years the number of the infected patients decreased due to the close collaboration of our department and hygiene-epidemiological and veterinarian department, as well as sanitary inspection which resulted with early discovery and location of the sources of infection and beginning of appropriate therapy.

P584 Hearing Loss in Brucellosis

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Objectives: Brucellosis is a disease of domestic and wild animals that is transmittable to humans. There is a wide spectrum of clinical signs and symptoms in brucellosis. Hearing loss is one of them but only few reports can be found in literature. This study was planned to show the importance of hearing loss in brucellosis.

Methods: In a period of 7 months, from January 1996 to July 1996, 83 patients were defined as brucellosis in Department of Infectious Diseases of Osmangazi University Hospital. According to other risk factors of hearing loss 23 of them were excluded from the study. The control group was including 41 patients submitting to hospital for different complaint which were negative for clinical and laboratory findings of brucellosis. Hearing loss was investigated by odiometric examination in the study and control groups.

Results: Hearing loss was identified in 28 (46.6%) cases of study group. There isn't a difference between male and female. In control group, hearing loss was identified in 8 (19.5%) cases ($p < 0.01$). Hearing loss was found more frequent in over 30 years old patients.

Conclusion: Since there was a few reports in literature about hearing loss in brucellosis, the results of this study suggest that it is an important manifestation of brucellosis and it will be useful to examine the hearing functions of patients with brucellosis especially in endemic areas.

P585 Clinical Characteristics and the Use of Magnetic Resonance Imaging (MRI) in Brucellar Sacroiliitis

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Objectives: To describe the clinical characteristics and the MRI of patients (p.) with brucellar sacroiliitis.

Methods: The clinical information and imaging studies from p. (examined 1989 through 1996) with the diagnosis of brucellar sacroiliitis were reviewed. The diagnosis was established on the basis of the following criteria: (1) one or more imaging examination with findings consistent with sacroiliitis; (2) isolation of *Brucella* spp from blood cultures (13 of 32 p.), or characteristics clinical findings and a standard tube agglutination titter of 1/160 or more (19 of 32 p.).

Results: Mean age was 22 (ranging from 8 to 73); 25 were male and 7 were female. The duration of symptoms before therapy ranged from 4 to 240 days (median, 41). All p. had gluteal and sacral pain and 81% experienced fever. Simultaneously, one p. presented spondylitis. The MRI showed a periarticular hypointensity lesion on T1W SE images and a hyperintensity lesion on T2W SE images which were enhanced after the administration of gadolinium. No periarticular abscesses were found. Four treatment groups were administered: group A, gentamicin for 7 days plus doxycycline for 30 or 45 days (n = 10); group B, doxycycline and rifampin for 45 days (n = 6); group C, netilmicin for 7 days plus doxycycline 45 days (n = 10); and group D, streptomycin for 14 days and doxycycline for 45 days (n = 6). 2 p. (6.2%) demonstrated therapeutic failure, one from group B and the other from group C. 2 other p. (6.2%) experienced relapse, one from group C and another from group D.

Conclusions: Brucellar sacroiliitis affects primarily young p., responding well to treatment. There are no differences in response to treatment among the different treatment groups used. MRI is useful in confirming the diagnosis of sacroiliitis.

P586 Osteoarticular involvement of brucellosis in Turkey

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No abstract available.

P587 Brucelosis Spondylitis

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Osteoarticularic system is the most frequently and often the most seriously endamaged during brucellosis. In the early phase of illness arthragra is expressed which can be associated with infectious syndrome it means that injuries are not always present, depending on phase and form of inflammation. As many authors state seizure of osteoarticularic system can be verified in 20 to 85% of all brucellosic patients. By precise analysis in 291 patients within 1991–1996. we have noticed high representation of osteoarticular pain as the symptom. Total 85.34% have had algic syndrome in anamnesis. The most of patients suffer pain in the lower part of back – 65.5%. Beyond this high percentage inflammation process is hidden and injuries can be found in the lower lumbal vertebrae and also in thorocal and cervic vertebrae. Finding of osteoarticularic damages is in direct dependance on two moments: the first is continuation of illness and the second one is sensitivity of instrumentarium.

Computer Tomography (CT) and Nuclear Magnetic Resonance (NMR) have been recently used to diagnoze these changes. We have used Röentgen diagnosis, Myelographia and Electromyelographia along with serological verification together with corresponding hygienic-dietetic regime.

P588 Molecular Epidemiology of Diphtheria in Algeria

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Objectives: Since 1993, more than 1000 cases of diphtheria were reported in Algeria. Strains of *Corynebacterium diphtheriae* (Cd) isolated from clinical cases were assayed by toxigenicity tests and typing methods.

Methods: A total of 70 Cd isolates collected randomly from 8 different regions of Algeria during the period 1993–1996 were characterized by toxigenic testing (Elek test or PCR), biotyping, antibiotyping and ribotyping.

Results: Ribotyping using *Bst*EII digestion revealed five ribotypes (B1 to B5) with a predominant pattern B1 which occurred for 60 of these isolates. The two ribotypes B3 and B4 were formed by a single strain. Use of *Pvu*II digestion allowed to further divide the ribotype B2 (n = 5) into two patterns which are correlated with their antibiotypes. The ribotypes B1 and B2 contain both biotypes *mitis* (n = 62) and *gravis* (n = 3) and both toxigenic (n = 59) and non-toxigenic strains (n = 6). No correlations were found between ribotypes and the geographic origin of patients. According to all the typing methods, we obtained 11 different types of Cd with a same type (*mitis*, tox +, resistant to minocyclin and chloramphenicol,

same ribotype *Bst*EII) for 54 of the 70 isolates. Unlike the Russian outbreak, the biotype *mitis* predomined (96%).

Conclusions: These results indicate that in analysing outbreaks of diphtheria, the use of genomic (i.e ribotyping with more than one enzyme) and phenotypic methods (biotyping, antibiotyping) allows to delineate more critically the Cd strains. Combination of these methods led to the identification of 11 different types but the presence of a predominant type for 77% of isolates confirms the explosive form of the outbreak of diphtheria in Algeria.

P589 Molecular Epidemiology of Non-Toxigenic *Corynebacterium Diphtheriae* Infection in England and Wales in 1995

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Objectives: To examine the molecular epidemiology of non-toxigenic strains of *Corynebacterium diphtheriae* from England and Wales in 1995 by ribotyping and PFGE. To compare the molecular types with data available from a comprehensive epidemiological questionnaire.

Methods: All 132 isolates of non-toxigenic *C. diphtheriae* referred from 50 labs to the Streptococcus and Diphtheria Reference Unit in 1995 were characterised by biotyping, toxigenicity testing and subjected to ribotyping. Representative strains were examined by PFGE. Questionnaires to obtain additional epidemiological and clinical data were sent to follow up all cases.

Results: 97% of isolates were from respiratory samples and most were pharyngitis cases. Of the 132 isolates, 104 (78.9%) belonged to the *gravis* biotype, 21 (15.9%) to *mitis* and 7 (5.3%) to *belfanti*. Ninety-six of 104 *gravis* biotypes (92.3%) belong to one ribotype pattern which was found in many geographical areas over England and Wales. PFGE of selected strains did not provide further discrimination of this *gravis* biotype. In contrast there was greater diversity amongst the 21 *mitis* biotypes, with 13 distinct ribotype patterns and no predominant type.

Conclusions: These results demonstrate that a potential clonal group of non-toxigenic *C. diphtheriae var. gravis* associated with sore throats in England and Wales may have emerged.

P590 Molecular Characterization of *Corynebacterium diphtheriae* from Northwestern Russia

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Objectives: To determine the molecular characteristics of *Corynebacterium diphtheriae* isolates from clinical cases and asymptomatic carriers in northwestern Russia, isolated during the current diphtheria epidemic.

Methods: A total of 20 *C. diphtheriae* isolates were biotyped and their toxigenicity was determined by Elek test and passive hemagglutination assay to quantify the toxin production. Pulsed field gel electrophoresis (PFGE) of *Sfi* I cleaved genomic DNA was carried out using CHEFF DRII system (BioRad).

Results: All the isolates were toxigenic with variable levels of the toxin production. Among the 18 isolates of gravis biotype, 17 produced the r1 PFGE pattern previously shown to be predominant in the current epidemic; 1 gravis and both mitis isolates had new and unique PFGE patterns different from those reported earlier by De Zoysa et al. in 1995.

Conclusion: Toxigenic *C. diphtheriae* of gravis biotype and r1 PFGE pattern is still prevailing in northwestern Russia. PFGE typing is useful for discrimination among the isolates and monitoring the spread of the diphtheria epidemic.

P591 PI Culiarities of Oral Pharynx Microbial Landscape in Patients with Diphtheria and Angina

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Objectives: 1) To study the relations between microflora composition and severity of an infections disease in patients with diphtheria; 2) To reveal an etimologic agent and choose tactics for antibacterial therapy for angina patients.

Methods: 33 patients with different forms of diphtheria and 195 with angina were subject to bacteriological test. Smears were obtained from the sites of pathologically changed mucosa to isolate obligatory representatives of autoflora, as well as conditionally pathogenic microorganisms.

Results: In 16 patients with diphtheria no disturbance of oral pharynx microbial landscape was found in 17 patients a wide range of conditionally pathogenic microorganisms were isolated. Comparative analysis of clinical and microbiological features seemed to find no relations between severity of the disease and the microflora character. In angina patients different bacterial pathogens and their associations were found. All of them formed a massive focus with a density of 10^5–10^7 CFU.

Conclusion: Staphylococcus epidermidis is considered to be a possible etiological agent if its density exceeds 10^5 CFU. A leading role of hemolytic Streptococcus and Staphylococcus aureus in angina etiology is determined.

P592 Characteristics of Diphtheria Pathogen in Belarus

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Objectives: To study biochemical properties and toxigenicity of *Corynebac-terium diphtheriae* (C.d.) in 1995–96 for the purposes of improvement of diphtheria surveillance in Belarus.

Methods: 428 *C.d.* strains, which were isolated from patients with diphtheria (70), contacts (98), healthy carriers (138), patients with sore throat (122), residing in all regions of the Republic were under studies.

Results: Pathogens of *gravis* and *mitis* biotypes were isolated in all groups of examined individuals. *Gravis* strains prevailed in diphtheria patients, contacts, healthy carriers and they made up 84.7%, 65.3%, 66.7% ($p < 0.05$), respectively. Toxigenic strains played a key role in diphtheria patients, among them *gravis* strains constituted 95.0%, *mitis* strains – 70.0%. In contacts toxigenic strains of gravis type made up 78.1%, but toxigenic strains of mitis type – only 29.4% ($p < 0.05$). Among strains, isolated from patients with sore throat and carriers, the non-toxigenic strains prevailed irrespective of their biotype (gravis – 60.4% and 57.6%; *mitis* – 95.7% and 93.5%, respectively; $p < 0.05$). Additionally, toxigenicity was investigated with PCR in 16 *C.d.* strains, isolated from diphtheria patients. Divergency of the data was revealed in one strain, that was non-toxigenic in Eleck test, but it carried toxigene A fragment.

Conclusions: Complex studies on properties of C.d. strains, circulating in Belarus, could be applied for the determination of pathogen origin, ways of its transmission, which are an object of special importance for diphtheria surveillance.

P593 Detection and Differentiation of *Leptospira* sp. by PCR Method

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Objectives: Detection of *Leptospira* sp. in clinical specimens. Differentiation of *Leptospira* serovars with (1) junction of two techniques: polymerase chain reaction (PCR) and restriction fragment length polymorphism (RFLP), and (2) fingerprinting polymerase chain reaction (FP-PCR).

Methods: Twenty seven cultures' of *Leptospira* serovars and *Leptospira icterohaemorrhagiae* serially diluted strains (in blood, sperm and urine) were tested by PCR with *Leptospira* genus-specific primers. Experimentally infected blood samples were examined by the same test. Nineteen *Leptospira* serovars were analysed by FP-PCR with L1 and G1 primers. The 16S rRNA of *Leptospira* serovars genes were amplified with PCR and analysed by RFLP with *AluI* enzyme. The DNA was extracted by kits made by A&A Biotechnology, Poland.

Results: The *Leptospira* DNA was detected in urine, sperm and blood. The PCR test enables detection of 10 *Leptospira* cells in PCR probe and was genus-specific for all tested samples. Total time of the test was less than 6 h. Leptospires can be detected in blood on second day of the infection. Both tests PCR-RFLP and FP-PCR gave partially differentiation of *Leptospira* serovars.

Conclusion: Test based on PCR technique is useful for quickly, sensitive and specific detection of leptospires in blood, sperm, urine, and give very early diagnosis of leptospirosis. Both differentiating tests may be used in epidemiological studies as alternative, additional method.

P594 Early Serodiagnosis of Serious Forms of Leptospirosis by IgM-ELISA

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Objectives: To study the epidemiological and clinical findings of leptospirosis in our geographical area, and to evaluate the utility IgM-ELISA test to detect leptospire-specific immunoglobulin M for early serodiagnosis of severe forms.

Patients and Methods: Fifteen patients with clinical diagnosis of leptospirosis were evaluated. A technique of indirect hemagglutination (IH) (*Hillcrest Biol.*) as test for screening was used. A title >1:50 was considered negative. To detect the IgM anti-leptospira specific of gender an ELISA (*PanBio*) was employed.

Results: From January 1993 to December 1996 the diagnosis of leptospirosis was made in 15 patients out of 86 patients with initial suspicion. Seroconversion or initial positive title of IH was detected in 8 patients. In 11 patients with initial negative or doubtful titles of IH, IgM ELISA was positive in 9 of them. Of 9 patients with initial IH negative or doubtful and IgM-positive, 5 of they had serious disease. From all patients eight suffered a serious leptospirosis and three died. Four patients needed admission in a intensive care unit. During the clinical course renal failure was observed in 7 cases, rabdomiolysis in 6, thrombopenia in 6, meningoencefalitis in 3 and shock with multiple organs failure in other 3.

Conclusions: Leptospirosis had a severe presentation in approximately 50% of the cases and a mortality rate of 20% of the global series. A high incidence of severe renal, pulmonary affectation, and rabdomiolysis was observed. The determination of IgM anti-leptospira by IgM-ELISA improves the efficiency of the usual serodiganosis by IH, mainly in the most serious forms of leptospirosis.

Lyme borreliosis

P595 Simultaneous Presence of Two *Borrelia burgdorferi sensu lato* Species in *Ixodes ricinus* Ticks

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Hematophagous arthropods, *Ixodes ricinus* ticks are transmitters of agent of Lyme disease *Borrelia burgdorferi* to men and animals. Recently it has been demonstrated that DNA from more than one of the three *Borrelia burgdorferi* sensu lato species was present *I. ricinus* nymphs (Pichon et al., Emerging Infect. Dis., 1995; 1 (3): 89–90). We analyzed 15 from 73 PCR positive for *Borrelia* DNA ticks (Jenek and Glazaczow, Przeg. Epid., 1996; 50: 383–386) for simultaneous presence of more than one genospecies in unfed adult ticks by using PCR amplification of flagelin gene fragment and hybridization with genospecies-specific probe.

Thirteen ticks were infected by a single species of *Borrelia* (eigth by *B. garinii* and five by *B. afzelii; Borrelia burgdorferi sensu stricto* was not found), and two were infected by both *B. garinii* and *B. afzelii.*

These preliminary data suggest that the simultaneous presence of more than one genospecies in arthropod vector may be not exceptional.

P596 *Borrelia burgdorferi sensu lato* DNA in Serum of Patients with Early Lyme Disease

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The studies aimed at evaluating diagnostic capacity of PCR technique and of serological techniques in *erythema chronicum migrans* (ECM) diagnosis.

Retrospective studies on *Borrelia burgdorferi* (*B.b.*) DNA presence in serum of patients with early borreliosis were conducted using PCR and hybridization with specific probe. Fifty four sera were tested earlier for presence of anti-*B.b.* antibodies (Enzygnost Borreliosis, Behring). High titre of specific anti-*B.b.* antibodies were found in 9 sera (IgG antibodies in 3 sera, IgM antibodies in 3 sera and in 3 sera both types of antibodies). In 7 cases specific activity of serum IgM was at the trace level. Presence of *B.b.* DNA was noted in 16 sera of which 5 only contained specific anti-*B.b.* antibodies.

The obtained results indicate that ECM diagnostics based on immunoserological techniques is insufficient and in clinically suspected cases should be supplemented by PCR studies.

P597 Lyme Disease as an Epidemiological Problem in Poland

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The first serological study in group of a high risk was carried out in Poland in 1991 (Anusz et al.). In 61 out of 417 persons (14.6%) antibodies against *Borrelia burgdorferi* were found.